Cannabidiol and Tetrahydrocannabinol concentrations in commercially available CBD E-liquids in Switzerland

Abstract

Cannabidiol (CBD) rich hemp and hemp products low in Δ^9-tetrahydrocannabinol (THC) (less than 1 %) are legally available in Switzerland. Besides herbs for smoking and oils, liquids (e-liquids) for smoking in electronic cigarettes (e-cigs) have recently appeared on the market. These e-liquids are available with different CBD concentrations and can be flavoured. The aim of the current study was to investigate 20 e-liquids legally available in Switzerland for their contents using Fourier-transform infrared spectroscopy (FTIR) as a preliminary step followed by gas-chromatography coupled to mass spectrometry to identify potential cannabinoids, natural plant compounds and flavours. Quantification of CBD, cannabidiol carboxylic acid (CBD-acid), cannabinol (CBN), Δ^9-tetrahydrocannabinol (THC), and Δ^9-tetrahydrocannabinol carboxylic acid A (THC-acid) was performed by a validated method with ultra-high-pressure-liquid chromatography coupled to a diode array detector (UHPLC-DAD).

FTIR analysis could confirm that for all investigated samples the e-liquid matrix consisted of 1,2-propanediol and glycerol. The qualitative GC-MS could identify ten phytocannabinoids including the quantified analytes, six natural plant compounds and five flavours.

All analysed samples had a total THC content below 0.1059 % (by weight), hence meeting the legal requirements of both Switzerland (< 1%) and the European Union (< 0.2%). The total CBD content ranged from 0.182 to 3.346 % and differed in ten out of 20 samples from the CBD content presented by the manufacturer by more than 10 % relative CBD. Furthermore, two of the analysed samples contained only 0.348 % and 0.182 % total CBD despite being labelled as "CBD rich". Seven of the 20 samples contained the correct CBD content (in the range of the labelled CBD content ± 10 %).

In conclusion, a deviation in the determined total CBD content from the labelled CBD content could be observed for half of the analysed samples, meaning that consumers cannot rely on the manufacturers' information. It is remarkable, that currently no official regulations for providing correct information of CBD content or any external product control is available in Switzerland and in most other countries.

Introduction

In Switzerland the legal status of cannabis is regulated by the Federal Act on Narcotics and Psychotropic Substances (BetmG 812.121). Cannabis is considered illegal if the total tetrahydrocannabinol (THC), the main psychoactive component of marihuana, content exceeds 1 %. However, Cannabidiol (CBD) rich hemp with a total THC content below 1 % can be legally obtained as a tobacco substitute product, CBD rich oils or in so called e-liquids [1, 2], which are mainly 1,2-propanediol (propylene glycol) and glycerol based (see Table 1).

Cannabis sativa contains 489 naturally compounds of which roughly 90 are phytocannabinoids [3, 4]. These substances are terpenophenols, which are mostly responsible for the plants psychoactive effects [3]. The main psychoactive compound of cannabis is Δ^9-tetrahydrocannabinol (Δ^9-THC), producing euphoria, relaxation analgesic, anti-inflammatory, appetite stimulating and antiemetic effects [5]. Yet, chronic Δ^9-THC consumption has been associated with severe side effects such as cognitive deficits, anxiety, paranoia, chronic psychosis and dependence [6-8]. CBD is a non-psychoactive isomer of Δ^9-THC and has anti-inflammatory, anticonvulsive, anxiolytic, analgesic, neuroprotective, anticancer and antioxidant effects [9].

Since 2006, vaping with electronic cigarettes (e-cigarettes) has become popular as an alternative to smoking cigarettes [10]. Different generations of e-cigarettes are available such as atomizers, cartomizers or clearomizers. Clearomizers are the most common consisting of a disposable head containing the heating coil and wicks converting the electronic liquids (e-liquids) into an aerosol (vapour), and a clear tank containing the e-liquid [11]. E-liquids commonly consist of 1,2-propanediol and vegetable glycerol and can additionally contain nicotine or different flavours (see Table 1). In Switzerland, recently, e-liquids containing Δ^9-THC or CBD have appeared on the market. Additionally, e-liquids containing synthetic cannabinoids have been reported to be found in Germany, Poland and in the USA [12-14].

A reason for the popularity of vaping could be that earlier publications claimed that the produced vape contains fewer harmful chemicals compared to ordinary cigarettes or joints [15, 16]. However, recent studies have shown that e-liquid vapour induces oxidative stress, glutathione depletion and increased production of inflammatory cytokines in human airway epithelial cells *in vitro* and in lungs of mice *in vivo* [10, 17]. Staudt et al. demonstrated in their recent publication that short-time vaping (two exposures to vapour of 10 puffs with a 30 min break) is harmful to the lung even in the absence of nicotine [18].

Recently, an increase of life-threating lung diseases (so called vaping-associated pulmonary illness (VAPI), over 1000 reported cases) and deaths (18 cases) has been reported in context with vaping of e-liquids in Northern America [19, 20]. The Centres for Disease Control and Prevention (CDC) suspects Δ^9-THC-containing and Δ^9-THC- and nicotine-containing products to be responsible for these adverse effects but is still investigating [19]. Lately, the recreational use of marihuana has become legal in eleven US states and another 15 US states have decriminalized its consumption [21]. In Switzerland two cases of patients with lung diseases in relation with e-cigarettes have been reported [22].

The aims of this study were to first investigate the composition of 20 different CBD rich e-liquids from Swiss headshops and a Swiss online retailer using Fourier-transform infrared spectroscopy (FTIR) and gas-chromatography mass spectrometry (GC-MS) to identify potential adulterations with synthetic cannabinoids, and second, to quantify the e-liquids' total Δ^9-THC and CBD concentrations with a previously published validated method with ultra-high pressure-liquid chromatography coupled to a diode-array-detector (UHPLC-DAD) [23], which we additionally validated for e-liquids.

Tables

Table 1: List of investigated E-liquids.

Sample number	Name	Manufacturer	Origin	Place of purchase	Date of purchase	CBD content Label	Flavour	Ingredients
1	Cannabis Botanical Dreams Mix	Merlins Garden	Switzerland	Werners head Shop Limmatquai 74 8001 Zurich	14.09.2019	not specified	Flavourless	not specified
2	Original Hemp	Marry Jane	Mary Jane GmbH Baarestrasse 141 6300 Zug, Switzerland	Marry Jane Niederdorfstrasse 58 8001 Zurich	14.09.2019	100 mg	Flavourless	not specified
3	Original Hemp	Marry Jane	Mary Jane GmbH Baarestrasse 141 6300 Zug, Switzerland	Marry Jane Niederdorfstrasse 58 8001 Zurich	14.09.2019	300 mg	Flavourless	not specified
4	Strawberry Wild	Harmony	London, UK	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen	17.09.2019	100 mg	Strawberry	propylene glycol, vegetable glycerine, aroma, CBD (methyl cinnamate, furaneol)
5	Kiwi Skunk	Harmony	London, UK	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	100 mg	Kiwi	propylene glycol, vegetable glycerine, aroma, CBD (Caryophyllen, D-limonen, linalool, β-pinen)
6	Exodus Cheese	Harmony	London, UK	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	100 mg	Cheese	vegetable glycerine, propylene glycol, aroma, CBD (caryophyllen, D-limonen, linalool, α-pinen, β-pinen)
7	Moroccan Mint	Harmony	London, UK	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	100 mg	Mint	vegetable glycerine, propylene glycol, aroma, CBD (carvone, D-limonen)
8	New-York Diesel	Harmony	London, UK	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	100 mg	Diesel	vegetable glycerine, propylene glycol, aroma, CBD (caryophyllen, D-limonen, linalool, β-pinen)

9	Original Hemp	Harmony	London, UK	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	100 mg	Flavourless	vegetable glycerine, propylene glycol, aroma, CBD
10	Cannabis Botanical Dreams- Zitrone	Merlins Garden	Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	not specified	Flavourless	60 % propylene glycol, 19 % herb extract, 17 % glycerine, 4 % dest. Water, 1% lemon oil
11	Cannabis Botanical Dreams-Orange	Merlins Garden	Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	not specified	Flavourless	60 % propylene glycol, 19 % herb extract, 17 % glycerin, 4 % dest. Water, 1% orange oil
12	Freedom	Cannaliz® Terpenes+	Cannatract Labs des Aux 14 1228 Plan-les-Quates Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	300 mg	not specified	45.5 %propylene glycol USP, 45.5 vegetable glycerine USP, 6 % Organic hemp flower cold-alcohol-extract, 3 % added terpenes (terpinolone, caryophyllene, myrcene, limonene), 0 % nicotin
13	Dreams	Cannaliz® Terpenes+	Cannatract Labs des Aux 14 1228 Plan-les-Quates Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	300 mg	not specified	45.5 % propylene glycol USP, 45.5 vegetable glycerine USP, 6 % Organic hemp flower cold-alcohol-extract, 3 % added terpenes (myrcene, caryophyllene, α-pinene, β-Pinene), 0 % Nikotin
14	Mojito	Cannaliz® Terpenes+	Cannatract Labs des Aux 14 1228 Plan-les-Quates Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	300 mg	Mojito	45.5 % propylene glycol USP, 45.5 vegetable glycerine USP, 6 % Organic hemp flower cold-alcohol-extract, 3 % added terpenes (linalol, caryophyllene, limonene, mentol), 0 % nicotin
15	Tangie	Swiss E-liquid/Pure Production	Pure Produciton Ag CBD-Shop hanfhof.ch Etzmatt 273, 4314 Zeinigen, Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	1%	not specified	propylene glycol, vegetable glycerine, natural aroma tangie, vegetable pure CBD 1 %

16	Amnesia	Swiss E-liquid/Pure Production	Pure Produciton Ag CBD-Shop hanfhof.ch Etzmatt 273, 4314 Zeinigen, Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	1%	not specified	propylene glycol, vegetable glycerine, natural aroma amnesia, vegetable pure CBD 1 %
17	Critical	Swiss E-liquid/Pure Production	Pure Produciton Ag CBD-Shop hanfhof.ch Etzmatt 273, 4314 Zeinigen, Switzerland	Hanfpost Wonderland Trading GmbH Kastelstrasse 89 2540 Grenchen, Switzerland	17.09.2019	1%	not specified	propylene glycol, vegetable glycerine, natural aroma critical, vegetable pure CBD 1 %
18	Lemon	Mary Jane	Mary Jane GmbH Baarestrasse 141 6300 Zug, Switzerland	Marry Jane Niederdorfstrasse 58 8001 Zurich	11.10.2019	1%	Lemon	propylene glycol, glycerine, CBD, aroma
19	Strawberry Wild	Mary Jane	Mary Jane GmbH Baarestrasse 141 6300 Zug, Switzerland	Marry Jane Niederdorfstrasse 58 8001 Zurich	11.10.2019	1%	Strawberry	glyercine E422, propylene glycol E1520, Cannabis Sativa L. ethanol extract
20	Melon	Mary Jane	Mary Jane GmbH Baarestrasse 141 6300 Zug, Switzerland	Marry Jane Niederdorfstrasse 58 8001 Zurich	11.10.2019	1%	Melon	glyercine E422, propylene glycol E1520, Cannabis Sativa L. ethanol extract

Methods

Chemicals and reagents

Ultrapure water was produced in-house using the direct-Q purification system from Millipore (Zug, Switzerland). Acetonitrile (HPLC gradient grade, 99.9 %) was purchased from Acros Organics (Chemie Brunschwig, Basel, Switzerland), n-hexane (EMSURE®) from Merck (Darmstadt, Germany), methanol (absolute, HPLC grade) from Biosolve (Chemie Brunschwig, Basel, Switzerland), formic acid solution (puriss p.a., 50 % in water), 1,2-propanediol (> 99.5 %) and glycerol (> 99.5%) from Sigma-Aldrich (Buchs, Switzerland). The reference standards of THC, tetrahydrocannabinolic acid A (THC-acid), CBD, cannabidiolic acid (CBD-acid) and cannabinol (CBN) were obtained from Lipomed (Arlesheim, Switzerland).

E-liquids

Twenty different e-liquids from five different manufacturers were purchased from two different headshops in Zurich, Switzerland and from the internet (www.hanfpost.ch) (compare Table 1). According their label, they contain 100-300 mg (1-3 %) of CBD with the exception of sample 1, 10 and 11 (Cannabis Botanical Dreams-Mix/ Zitrone/ Orange), which had no specification. Fourteen e-liquids were produced in Switzerland and six in London, UK (manufacturer Harmony) according to their label. Six e-liquids were unflavoured (sample 1-3, 9-11) and 14 were flavoured (sample 4-8, 12-20).

Sample preparation

For the qualitative GC-MS analysis of the e-liquids, 200 µL e-liquid were transferred into a polypropylene vial and extracted with 1000 µL of a mixture of n-hexane/ ethyl acetate (7:3, v/v). Samples were shaken for five minutes followed by centrifugation for 10 min at 8 °C and 17,000 g. The supernatant was transferred into a new vial and an aliquot of 10 µL was diluted with 990 µL ethyl acetate.

Sample preparation for the quantification of CBD, CBD-acid, CBN, THC and THC-acid using UHPLC-DAD was as follows: 150 mg of e-liquid were diluted with 10 mL of a mixture of methanol/ n-hexane (9:1, v/v). Aliquots were diluted 1:10 (v/v) and 1:20 (v/v) with the methanol/n-hexane mixture and transferred to another autosampler vial, respectively. To measure the low CBD-acid, CBN, THC and THC-acid concentrations, 150 mg e-liquid were diluted with 1 mL methanol/n-hexane mixture, shaken, ultrasonicated and directly transferred to an autosampler vial for analysis. Samples were analysed in duplicates.

Qualitative analysis

FTIR and GC-MS were used for qualitative analysis. Our laboratory has been accreditated according to ISO 17025:2018 in 2019 for Forensic Chemistry – including these two methods for qualitative analysis. The validation included determination of specificity and selectivity as well as sensitivity for a selection of drugs including synthetic cannabinoids.

Qualitative analysis using Fourier-transform infrared spectroscopy (FTIR)

In order to identify the solvent of the tested e-liquids an FTIR spectrometer with a Universal-Attenuated-Total-Reflectance polarization accessory (FTIR-UATR) and Spectrum v2.00 software (Perkin Elmer, Schwerzenbach, Switzerland) was used. Scan range was from 4000 cm^{-1} to 400 cm^{-1} with a resolution of 4 cm^{-1}. The liquid samples were directly analyzed without any pretreatment. For identification the following libraries were used: Drugs (Perkin Elmer, Massachusetts, USA), BAG Bern (Ministry of Health, Bern, Switzerland), Fluka (Perkin Elmer, Massachusetts, USA).

Qualitative analysis using GC-MS with electron impact ionisation (EI)

The e-liquids were qualitatively analysed using a 6890N gas chromatograph with a 7683B autosampler coupled to a 5973 inert mass spectrometer (Agilent, Basel, Switzerland). Chromatographic separation was performed on a 5 % phenyl methyl polysiloxane capillary column (30 m, 0.25 mm i.d., film thickness 0.25 µm; Perkin Elmer, Schwerzenbach, Switzerland). The total run time was 30.5 min and a temperature gradient was applied: 0-3 min: 80 °C, 3-7 min: 20 °C/min to 150 °C, 7-22 min: 10 °C/min to 300 °C, held at 300 °C for 8.5 min. Helium gas was used as carrier gas with a flow rate of 1 mL/min. The injection volume was set to 1 µL (splitless) and the solvent delay was 3 min. EI mass spectra were aquired with a 70 eV ionisation energy. The mass spectrometric scan range was from *m/z* 25 to 700 with a scan time of 1 sec/scan in the time window of 3.1 to 30 min.

Glycerol and 1,2-propanediol were confirmed by full-scan GC-MS analysis in EI-mode (same instrumentation) using a modified gradient: 0 – 2 min: 35 °C; 2 min – 3 min: 20°C/min to 55°C, 3 min: 40°C/min to 310°C, hold time: 8 min, with split-injection (split ratio 1: 30) of a 1.5mg/mL solution of the e-liquid in methanol/hexane (99:1, v/v). Retention times and characteristic masses (1,2-propanediol: 3.7 min; m/z 45, 43, 61; glycerol: 5.6 min; m/z 61, 43, 42).

For compound identification the mass spectral libraries "Wiley Registry of Mass Spectral Data with NIST" and "SWGDRUG MS Library" Version 3.5.L (September 23, 2019) were used.

The chromatograms and mass spectra were reproduced using GraphPad Prism 8.1.0.

Quantitative analysis using UHPLC-DAD

The quantification of CBD, CBD-acid, CBN, THC and THC-acid was carried out according to a previously published method [23] with an UltiMate 3000 UHPLC system (Dionex, Olten Switzerland) consisting of a HPG-3400RS binary pump, a WPS-3000TRS autosampler, a TCC-3000RS column compartment coupled to a DAD-3000RS detector with Chromeleon software Version 6.8 (Thermo Scientific, Rheinach, Switzerland). Chromatographic separation was performed with a Kinetex C8 column, 2.6 mm, 100 Å, 100 µm x 2.1 mm (Phenomenex, Basel, Switzerland) with the mobile phase consisting of water with 0.1 % formic acid (A) and acetonitrile with 0.1 % formic acid (B) and a flow rate of 0.6 mL/min. The following gradient was applied: 0–2 min: 50% B; 2–9 min: 50 % to 65% B; 9–10 min: 65% B; 10–10.1 min: 65 % to 50% B; 10.1–13 min: 50% B. The temperature of the autosampler and column compartment were set to 8 °C and 25 °C, respectively. The injection volume was 5 µL. The detection wavelength for quantification was 210 nm.

A six-point calibration was prepared in methanol for THC, CBD and CBN (0.001, 0.005, 0.01, 0.02, 0.05 and 0.1 mg/mL) and a seven-point calibration in methanol for CBD-acid and THC-acid (0.01, 0.05, 0.1, 0.2, 0.3, 0.4 and 0.5 mg/mL), respectively as described previously [23]. To obtain the total concentrations of CBD and THC, respectively, the concentrations of CBD and CBD-acid (or THC and THC-acid) are added up by using molar concentrations and then the sum of molar concentrations is recalculated as total CBD (or as total THC) concentration in g/100 g (weight %), thus compensating for a theoretical loss of CO_2. This total CBD-concentration (or total THC-concentration) is used for reporting the total CBD or total THC content in the e-liquids. The conversion factor of CBD-acid to CBD – as well as for THC-acid to THC - is 0.88 (MW_{CBD} / $MW_{CBD\text{-}acid}$ = 314.47 g mol^{-1} / 358.48 g mol^{-1}).

For the calibration curves the analyte concentration (x) was plotted against the peak area by linear least-squares regression using a $1/x^2$ weighted factor. When the correlation coefficient (R^2) is greater than 0.99, and the back-calculated calibrator concentrations were within ± 15% of the target value, linearity was acceptable. In order to determine selectivity of the method, the peak purity as well as the resolution of the analyte peaks from neighbouring peaks were assessed. Furthermore, the limits of detection (LODs) were defined as the concentration with a signal-to-noise ratio (S/N) of at least 3 by evaluation of the chromatograms of methanolic solutions containing decreasing concentrations of

analytes. The lower limit of quantification (LLOQ) was defined as the lowest calibrator of the respective calibration curve (THC, CBD and CBN 0.001 mg/mL, CBD-acid and THC-acid 0.01 mg/mL). In order to access precision two authentic samples and two samples consisting of spiked matrix in low and high concentration (0.4 % and 4%) were measured in sextuplets. Intra-assay precision was determined from six replicates in a single run. Spiked samples consisted of 1,2-propanediol and glycerol (70:30, w/w) and were spiked with methanolic solutions containing CBD, THC, CBN, CBD-acid and THC-acid to obtain a final concentration of cannabinoids of 0.4 % (sample A) or 4 % (sample B), respectively. Authentic and spiked samples were worked up as described earlier. Precision was expressed as percent relative standard deviation (%RSD) and was expected to be ± 20%RSD. Carry-over was assessed in six independent runs by injection of methanol/n-hexane (9:1, v/v) directly after the highest calibrators. Absence of carry-over was proven if the analyte peak in the solvent sample was lower than 20% of the LLOQ response. The chromatograms were reproduced using GraphPad Prism 8.1.0.

Results and Discussion

Twenty different CBD rich e-liquids from two different headshops and from the internet were qualitatively and quantitatively analysed using FTIR, GC-MS and UHPLC-DAD.

Qualitative analysis

Propylene glycol (1,2 propanediol) and glycerol (glycerine, propane-1,2,3-triol) were the best hits found by library search for the major solvents of the e-liquids by FTIR (Figure 1) and GC-MS (EI mode) (Figure 2) in all samples.

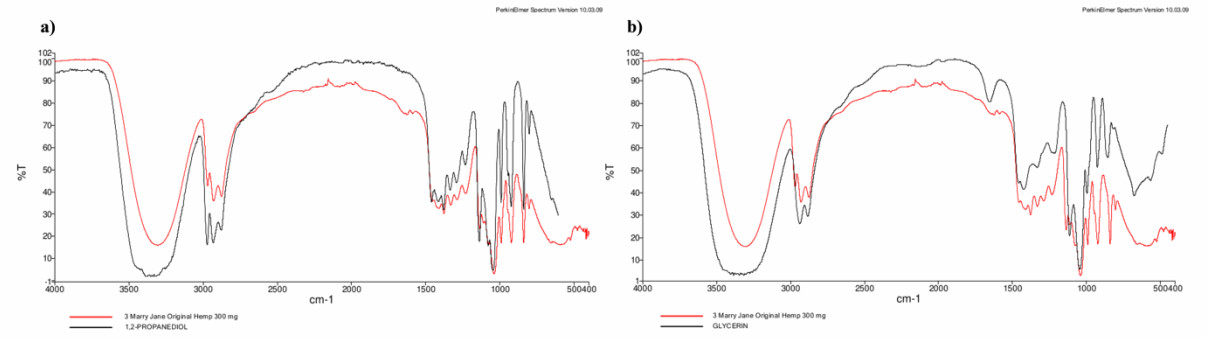

Figure 1: FTIR spectra of sample 3 (red, upper spectra) and library matches of a) 1,2-propanediol (propylene glycol) b) and glycerol acquired in the range of 4000 cm^{-1} to 400 cm^{-1}. Hits by library search for 1,2-propanediol and glycerol with a correlation factor of 0.8854 and 0.6376, respectively.

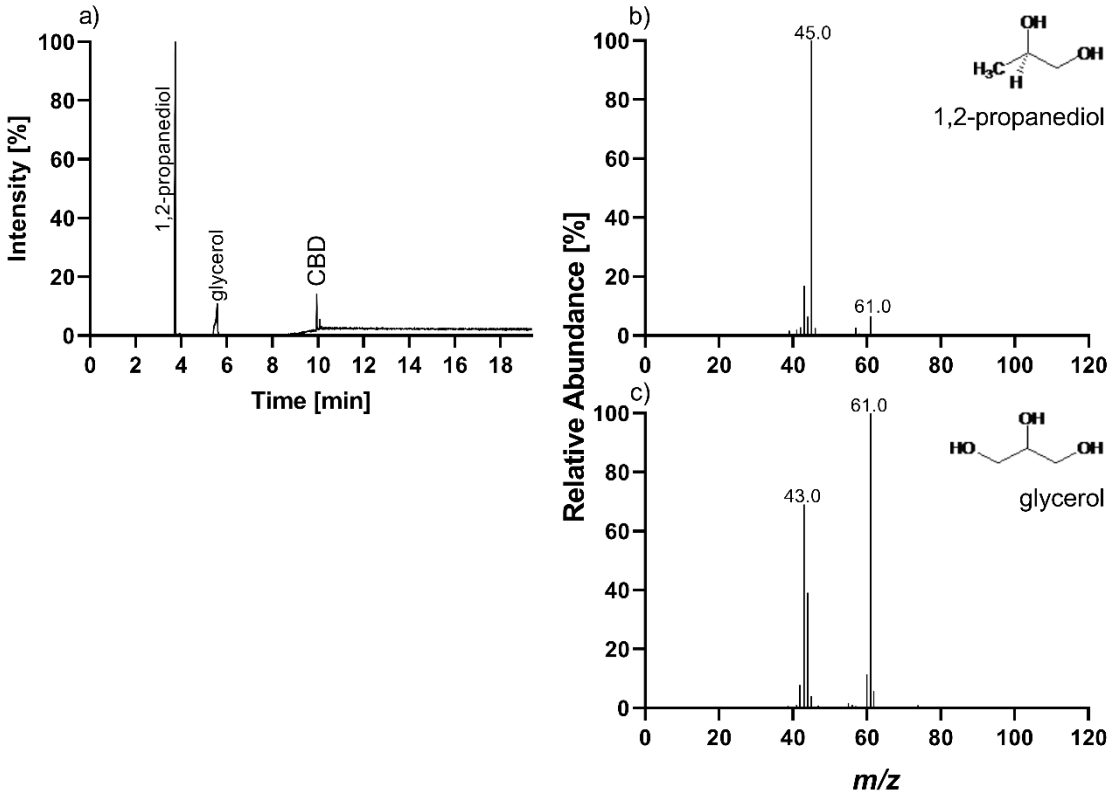

Figure 2: GC-MS (EI mode): a) representative total-ion chromatogram of e-liquid matrix of sample 13 and the corresponding mass spectra of b) 1,2-propanediol and c) glycerol.

In total ten different phytocannabinoids were identified by GC-MS, namely: CBD, CBN, Δ^9-THC, Δ^8-THC, Cannabicitran, Cannabielsoic acid, Cannabicyclol, Cannabidivarin, Cannabigerol, and Cannabichromene (see Figure 3 and Table 2). In all 20 samples the presence of CBD was confirmed and in 18 samples (samples 1-9 and 12-20) Δ^9-THC could be found. Cannabicitran and cannbielsoic acid were present in 15 samples, respectively. Example mass spectra are depicted in Figure 4.

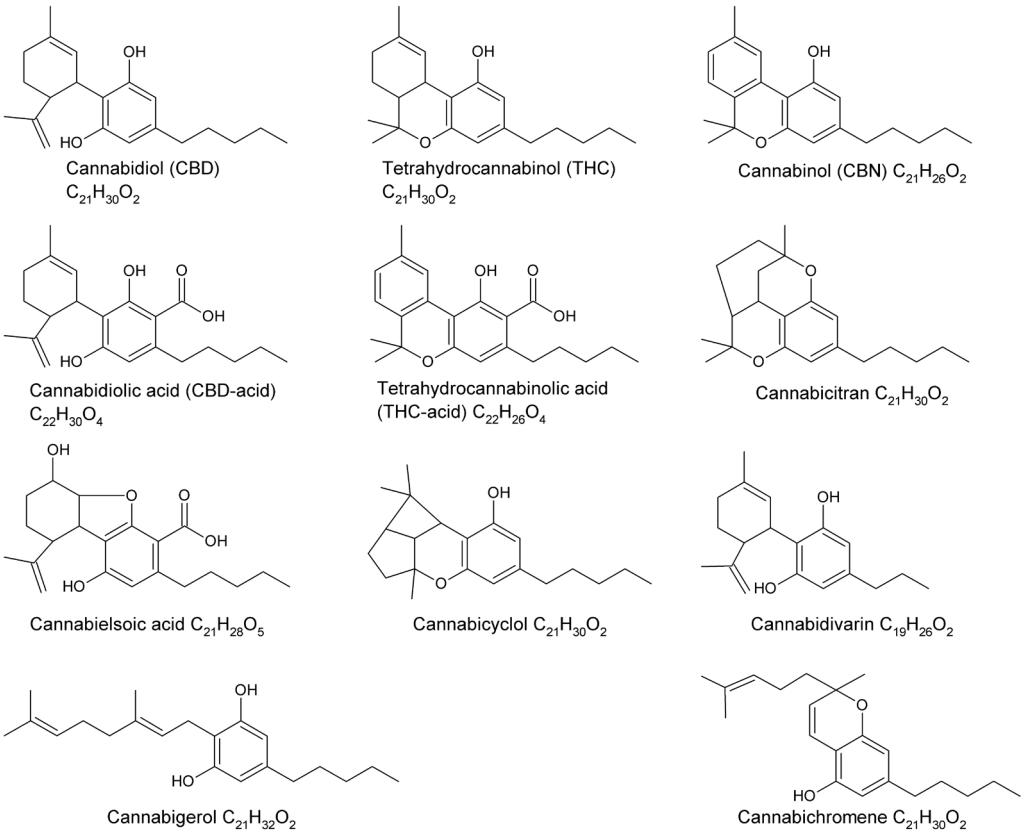

Figure 3: Chemical structures with their molecular formulas of the five phytocannabinoids quantified by UHPLC and the five additionally phytocannabinoids, which have been identified by GC-MS.

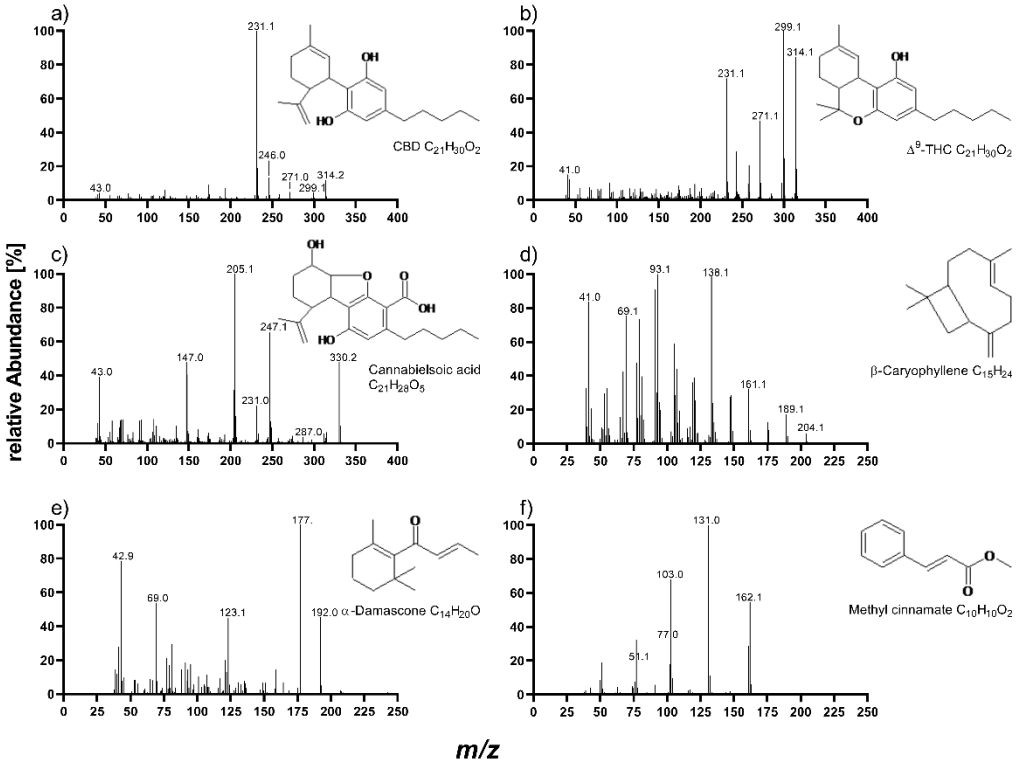

Figure 4: Mass spectra of three of the identified phytocannabinoids a) CBD, b) Δ9-THC, c) cannabielsoic acid, one additive d) β-caryophyllene, and two flavours e) α-damascone and f) methyl cinnamate.

Table 2: Identified phytocannabinoids in the respective e-liquid using GC-MS. +: detected and identified by GC-MS; -: not detected

Sample	CBD	CBN	Δ^9-THC	Δ^8-THC	Cannabicitran	Cannabielsoic acid	Cannabicyclol	Cannabidivarin	Cannabigerol	Cannabichromene
1	+	-	+	+	+	-	-	-	-	-
2	+	-	+	-	+	+	-	-	-	-
3	+	-	+	+	+	+	+	-	-	-
4	+	-	+	+	+	-	-	+	-	-
5	+	-	+	-	+	+	-	+	-	-
6	+	-	+	+	+	+	-	-	-	-
7	+	-	+	-	+	+	-	+	-	-
8	+	-	+	-	-	+	+	+	-	-
9	+	-	+	+	+	+	-	-	-	-
10	+	-	-	-	-	-	-	-	-	-
11	+	-	-	-	-	-	-	-	-	-
12	+	+	+	+	+	+	+	+	+	-
13	+	+	+	-	+	+	-	+	+	+
14	+	+	+	-	+	+	-	+	+	+
15	+	-	+	-	+	+	-	-	-	-
16	+	-	+	+	+	-	+	-	-	-
17	+	-	+	-	+	+	-	-	-	-
18	+	-	+	-	-	+	-	-	-	-
19	+	-	+	-	+	+	-	-	-	-
20	+	-	+	-	-	+	-	-	-	-

Besides the ten phytocannabinoids further six natural plant compounds (squalene, α-caryophyllene, β-caryophyllene, caryophyllene oxide, nerolidol) and five flavours (methyl cinnamate, α-damascone, monomethyl succinate, γ-decalacton, hedione and rasperry ketone) were identified by GC-MS (compare Figure 5 and Table 3).

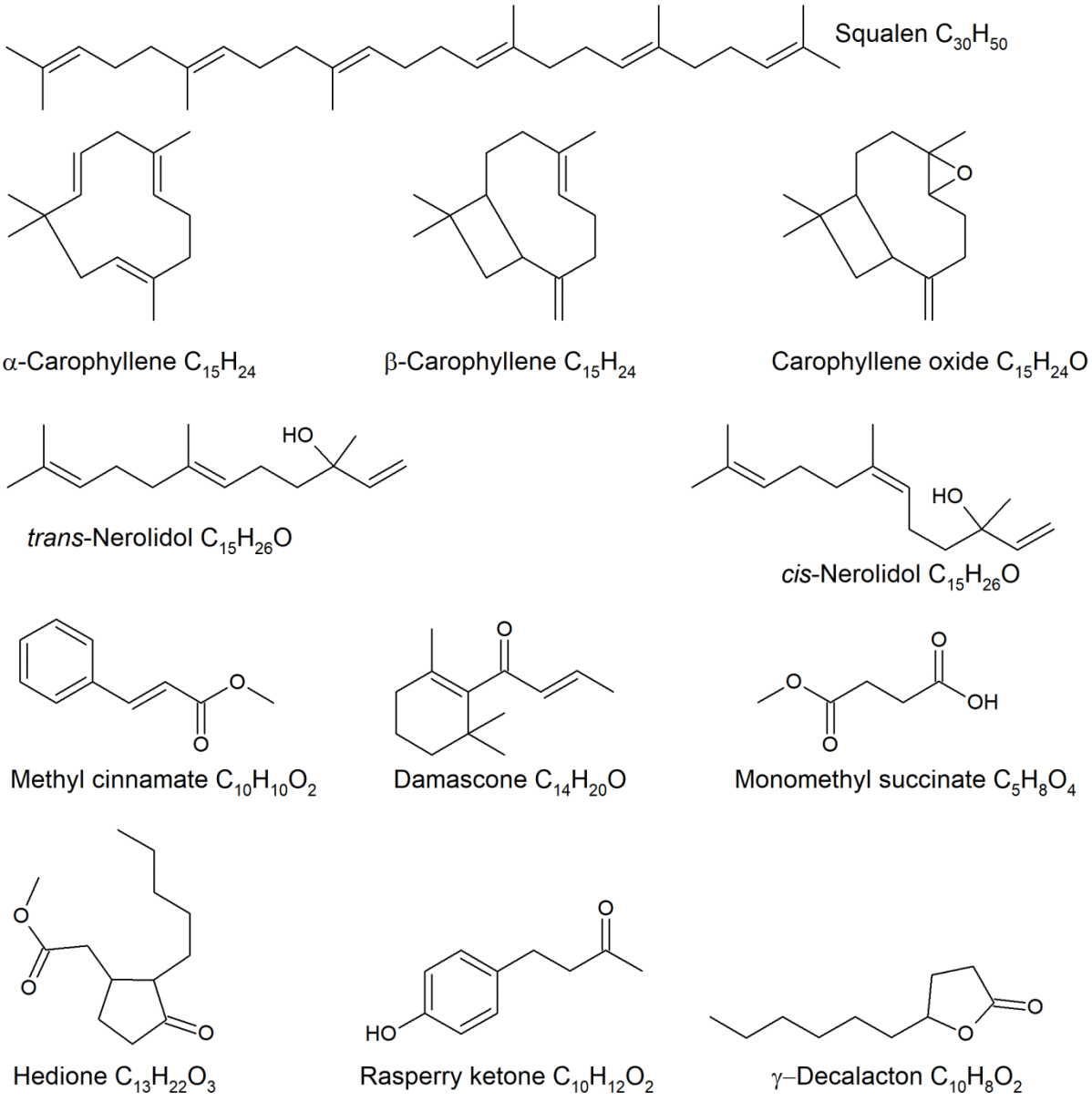

Figure 5: Chemical structures and their molecular formulas of additives and flavours in the 20 analysed e-liquids identified by GC-MS.

Table 3: Flavouring agents and other compounds in the 20 analysed e-liquids identified by GC-MS. +: detected and identified; -: not detected

Sample	Squalene	α-Caryophyllene	β-Caryophyllene	Caryophyllene oxide	*cis*-Nerolidol	*trans*-Nerolidol	Methyl cinnamate	α-Damascone	Monomethyl succinate	γ-decalacton	Hedione	Rasperry ketone
1	-	-	-	-	-	-	-	-	-	-	-	-
2	-	-	-	-	-	-	-	-	-	-	-	-
3	-	-	-	-	-	-	-	-	-	-	-	-
4	-	-	-	-	-	-	+	+	+	+	+	+
5	+	+	+	+	+	+	-	-	-	-	-	-
6	-	+	+	+	-	+	-	-	-	-	-	-
7	-	-	-	-	-	-	-	-	-	-	-	-
8	-	+	+	+	-	+	-	-	-	-	-	-
9	-	+	+	+	-	-	-	-	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-
11	-	-	-	-	-	-	-	-	-	-	-	-
12	-	+	+	+	-	-	-	-	-	-	-	-
13	-	+	+	-	-	-	-	-	-	-	-	-
14	-	+	+	-	-	+	-	-	-	-	-	-
15	-	+	+	+	-	+	-	-	-	-	-	-
16	-	+	+	-	-	-	-	-	-	-	-	-
17	-	+	+	-	-	-	-	-	-	-	-	-
18	-	-	-	-	-	-	-	-	-	-	-	-
19	-	-	-	-	-	-	-	-	-	-	-	-
20	-	-	-	-	-	-	-	-	-	-	-	-

Caryophyllenes were found in ten samples. Even though β-caryophyllene targets selectively the CB_2 receptor and has anti-inflammatory properties, it is not a phytocannabinoid but a sequiterpene [24]. There are two isomeric forms of caryophyllenes: α-caryophyllene (also known as humulene) and β-caryophyllene. They are often found in combination with α-caryophyllene or their oxidation product carophyllene oxide in essential oils of different spices such as oregano (*Origanum vulgar L.*), cinnamon (*Cinnamomum spp*.) and also *Cannabis sativa* [25-27]. Nerolidol was found in five samples and is a naturally occurring sesquiterpene alcohol. In nature, it can be found in various plants with a floral odour [28]. It is an additive in cosmetic and non-cosmetic products and as a flavouring agent [29]. Nerolidol has pharmacological and biological activities such as anti-microbial [30], anti-parasitic [31], anti-biofilm [32], anti-oxidant [33], anti-nociceptive [34], anti-inflammatory [34], anti-ulcer [35], skin penetration enhancing [36], and anti-cancer properties [29]. Nerolidol can be isolated from natural sources and via a chemical reaction were linalool is the starting reactant [29]. Interestingly, the ingredient lists of sample 5, 6 and 8 stated linalool as an ingredient. However, we could only identify nerolidol.

The six identified flavours were found in sample 4 (Figure 6), which was labelled with strawberry flavour and had the strongest odour out of all tested e-liquids. However, sample 4 did not contain any of the other additives.

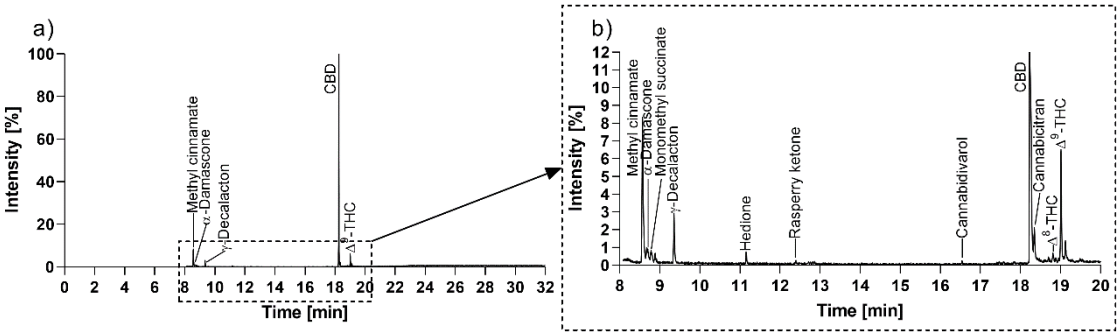

Figure 6: a) GC-MS (EI mode): Total ion chromatogram of sample 4. b) Heightened chromatogram from 8 to 20 minutes, peaks labelled with the identified compounds. The six identified flavours were eluting between 8 and 13 minutes and the four phytocannabinoids between 16 and 19 minutes.

Interestingly, sample 10 and 11 only contained CBD and neither any other phytocannabinoid nor additive. Presumably, this product was not made from plant extracts but from pure synthetic CBD, which is also available from internet shops. Sample 1-3 and 18-20 contained phytocannabinoids but no additives. Figure 7 displays the total ion chromatogram of sample 13, which contained eight different phytocannabinoids and two additives. The mass spectra of CBD, Δ^9-THC, β-caryophyllene, cannabielsoic acid (sample 13), α-damascone and methyl cinnamate (sample 4) are depicted in Figure 4.

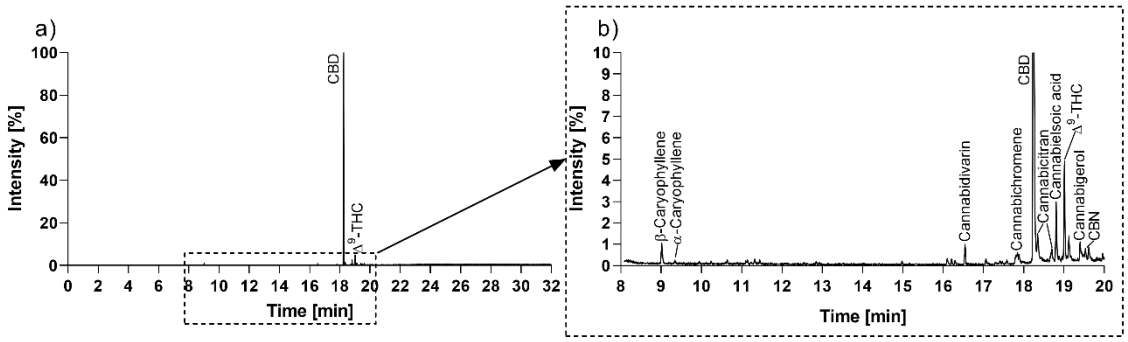

Figure 7: a) Total ion chromatogram of sample 13. b) Heightened chromatogram from 8 to 20 minutes labelled with the identified peaks. Eight different phytocannabinoids, eluting between 16 and 20 minutes, and two additives, eluting at 9 minutes, could be identified.

Quantitative analysis

The quantification results are depicted in Table 7. The total CBD concentration could be quantified in all 20 samples and was in a range from 0.18 % (sample 11) to 3.35 % (sample 3).

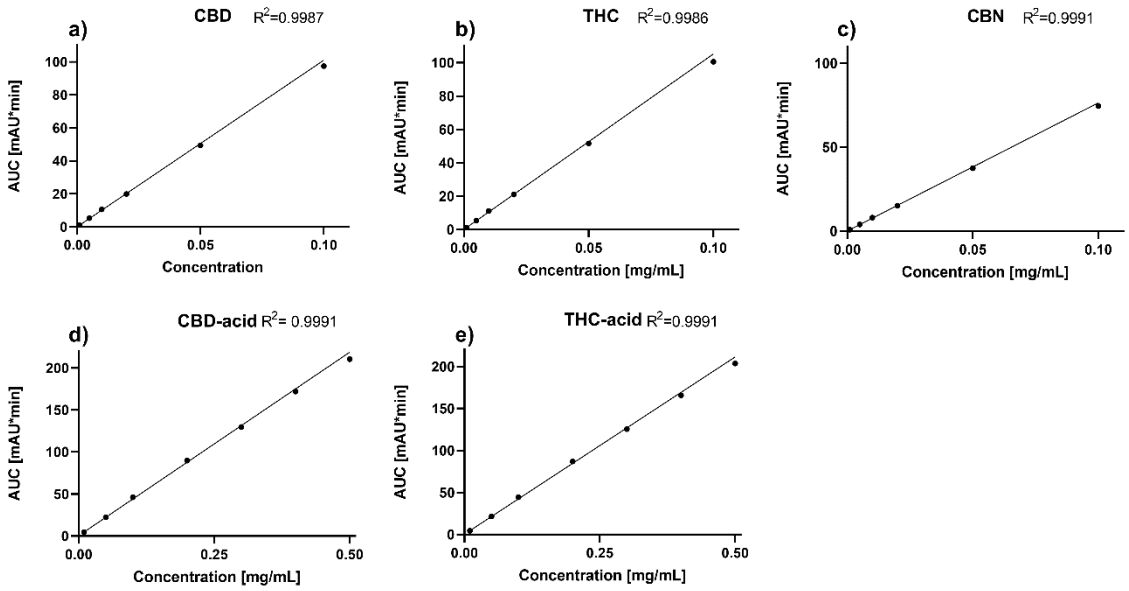

Figure 8: Calibration curves of CBD (a), THC (b), CBN (c), CBD-A (d) and THC-A (e) plotting the area under the curve (AUC [mAU*min]) versus the concentration [mg/mL], using weighted ($1/x^2$) linear regression.

The five analyte retention times were stable over a large number of injections and highly reproducible. The relative standard deviation (%RSD) of the retention time of all analytes in the quality control and precision samples of six runs was below 0.43 % (see Table 4). Representative chromatograms are depicted in Figure 8. The method showed linearity from 0.001, to 0.1 mg/mL for THC, CBD and CBN and from 0.01 to 0.5 mg/mL for CBD-acid and THC-acid, having correlation coefficients R^2 above 0.9986 for all analytes, using a weighted ($1/x^2$) linear regression (see Figure 8). The LODs were assessed on visual examination of the chromatograms (S/N≥3) and the LLOQ were defined as the lowest calibration standard (S/N≥10).

Table 4: Relative standard deviation (%RSD) of the retention times of all analytes in Sample A and the quality control samples of six runs.

Analyte	Retention time [min] n=18	standard deviation	%RSD
CBD-acid	5.68	0.02	0.410
CBD	6.26	0.02	0.349
CBN	7.47	0.02	0.295
THC	8.26	0.02	0.270
THC-acid	9.23	0.04	0.426

Carry-over was determined by injecting six times pure solvent (methanol/n-hexane, 9:1, v/v) after the highest calibrators (THC, CBD and CBN 0.1 mg/mL and CBD-acid and THC-aid 0.5 mg/mL). No analyte peaks were present in the pure solvents, hence, no carry-over took place.

For the assessment of accuracy two samples consisting of matrix (propylene glycol and glycerol) spiked with low (0.4 %) and high (4 %) concentrations of all analytes were measured in sextuplets. The accuracy was acceptable with a bias of -12.7 % to + 19 % (for the low concentrations) and -13.5 % to –7.9% for the high concentrations, respectively. Recoveries were from 87.3 % to 119 % in the low concentration range and from 86.5 % to 92.1 % in the high concentration range (compare Table 5). For the precision, additionally the two quality control samples (QC-6 and QC-11) were measured in sextuplets. The %RSD was between 0.22 % and 6.86 % and therefore acceptable (compare Table 6).

Table 5: Recovery data for e-liquid matrix (propylene glycol and glycerol) spiked with CBD, CBD-acid, CBN, THC and THC-acid.

Sample	Analyte	Spiked [%]	Found [%] n=6	Recovery [%]
A	CBD	0.40	0.47	119
	CBD-acid	0.40	0.39	97.5
	CBN	0.40	0.47	116
	THC	0.40	0.47	116
	THC-acid	0.40	0.35	87.3
B	CBD	4.0	3.7	92.1
	CBD-acid	4.0	3.5	88.1
	CBN	4.0	3.5	86.5
	THC	4.0	3.5	88.4
	THC-acid	4.0	3.5	87.6

Table 6: Inter-assay precision data for CBD, CBD-acid, CBN, THC and THC-acid for A (sample spiked with 0.4 % analyte concentration), B (sample spiked with 4 % analyte concentration), QC-6 quality control samples spiked with CBD, CBN and THC and QC-11 quality control sample spiked with CBD-acid and THC-acid. .

Sample	Analyte	Content [%] n=6	%RSD n=6
A	CBD	0.47	6.45
	CBD-acid	0.39	6.86
	CBN	0.47	4.51
	THC	0.47	4.82
	THC-acid	0.35	6.66
B	CBD	3.69	5.90
	CBD-acid	3.53	5.86

	CBN	3.46	5.45
	THC	3.54	5.37
	THC-acid	3.51	5.58
QC-6	CBD	0.11	0.32
	CBN	0.10	0.40
	THC	0.10	0.37
QC-11	CBD-acid	0.30	0.27
	THC-acid	0.30	0.22

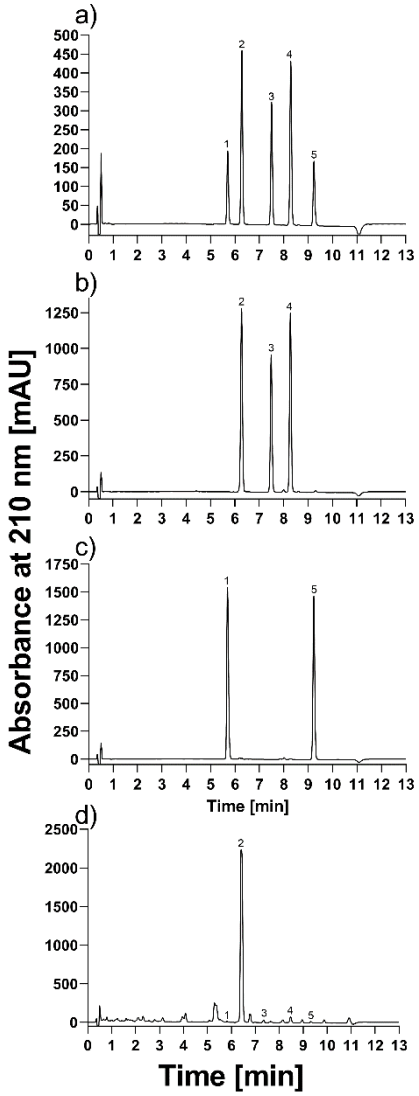

Figure 9: Representative HPLC chromatograms of a) cannabinoid standards in pure solvent at 0.4 % analyte concentration, b) a quality control with THC, CBD and CBN (QC-6) and c) a quality control with CBD-acid and THC-acid (QC-11) and d) an authentic e-liquid (sample 12). Peak assignment: 1. CBD-acid, 2. CBD, 3. CBN, 4. THC, 5. THC-acid.

Table 7: Concentrations of CBD, CBD-A, CBN, THC and THC-A in the 20 tested E-liquids. For CBD and THC concentrations are given in both mg/mL and % (w/w).

Sample	CBD content label	total CBD [%] (w/w)	rel. CBD content[1] [%]	CBD [%] (w/w)	CBD-acid [%] (w/w)	CBN [%] (w/w)	THC [%] (w/w)	THC-acid [%] (w/w)	total THC [%] (w/w)
1	not specified	1.421	-	1.420	0.00120		0.0056		0.0057
2	100 mg	0.858	85.8	0.858					
3	300 mg	3.346	111.5	3.346		0.0010			
4	100 mg	1.005	100.5	1.005			0.0011		0.0010
5	100 mg	0.881	88.1	0.881		0.0014	0.0011		0.0009
6	100 mg	0.817	81.7	0.817		0.0014		0.0011	0.0010
7	100 mg	0.889	88.9	0.889			0.0007		0.0006
8	100 mg	0.928	92.8	0.928		0.0009	0.0004	0.0005	0.0007
9	300 mg	3.009	100.3	3.009		0.0008	0.0069		0.0071
10	not specified	0.348	-	0.348			0.0014		0.0013
11	not specified	0.182	-	0.182			0.0011		0.0010
12	300 mg	2.787	92.9	2.778	0.0102	0.0230	0.0686	0.0427	0.1059
13	300 mg	1.376	45.9	1.352	0.0273	0.0106	0.0100	0.0118	0.0208
14	300 mg	2.296	76.5	2.280	0.0187	0.0242	0.0362	0.0289	0.0637
15	1%	0.899	89.9	0.872	0.0307			0.0004	0.0003
16	1%	1.060	106.0	1.022	0.0431		0.0013		0.0012
17	1%	1.179	117.9	0.980	0.2264		0.0012		0.0011
18	1%	0.999	99.9	0.999			0.0008		0.0006
19	1%	0.816	81.6	0.816					
20	1%	0.953	95.3	0.953			0.0017		0.0017

[1] ratio of total determined CBD content and labeled CBD content in %

For all 20 tested e-liquids the total THC content was lower than 1 %, meaning no legal violations. In fact the total THC concentration was below 0.2 % in all samples, which is the legal cut-off in Germany and the European Union for THC containing hemp products [37]. CBD-acid, CBN and THC-acid could be quantified in seven, seven and six samples respectively. A discrepancy between manufacturer information on the label and the measured total CBD concentration, which we define here as "rel. CBD content", was observed in a range from 45.9 % (sample 13) to 117.9 % (sample 17). A deviation from the labelled CBD content in the range of ± 10 % (relative) was set as acceptable. Sample 1, 10 and 11 were from the same Swiss manufacturer and had no CBD content specification but were labelled "CBD rich". For sample 1, a total CBD content of 1.421 % was determined. However, the two flavoured e-liquids (sample 10 and 11) only contained 0.348 % and 0.182 % total CBD, respectively. It is questionable if such a low total CBD concentration can have any effect. Sample 13 and 14 from another Swiss manufacturer were labelled with a CBD content of 300 mg and contained lower concentrations (rel. CBD content 45.9 % and 76.5 %, respectively). These two samples and sample 12 (rel. CBD content 93.9 %) from the same manufacturer also contained CBD-acid, CBN, THC and THC-acid and had the highest total THC concentrations (0.1059 %, 0.0208 % and 0.0637 %, sample 12, 13, 14, respectively) of all tested samples. It is surprising that products from the same producer have such a varying relative CBD concentration. The samples of the only non-Swiss producer (from the UK, samples 4-9) also showed variations in the relative CBD content (rel. CBD content 81.7 %- 100.3 %), however also had two samples (sample 4 and 9) with the most accurate total CBD concentrations.

Discussion

Ten out of 20 analysed e-liquids had a discrepancy between the labelled CBD content and the determined total CBD concentration of greater than ± 10 % (relative). Of those ten e-liquids, two had a higher and eight a lower total CBD concentration than labelled, respectively. Furthermore, the determined total THC content was in all samples lower than 0.1059 %, hence below the legal threshold of 0.2 % in Switzerland. Peace et al. [38] analysed two commercially available e-liquids for their CBD content and potential additives. The two analysed samples had a relative CBD content of 197 % and 230 % compared to the labelled content. No further cannabinoid or natural plant compound could be detected in the samples but 11 additives. Bonn-Miller et. al [39] investigated different CBD rich products (n=84) including 24 CBD rich e-liquids. They found that 12.5 % of the analysed e-liquids were accurately labelled, 75 % had a higher CBD content than labelled and 12.5 % had a lower CBD content than labelled. THC was detected (up to 6.43 mg/mL) in 18 of the 84 samples tested. Comparable to our results, they also observed that unlabelled products contained in general a very low CBD concentration [39]. Pavlovic et. al [40] investigated 15 CBD-based oil preparations commercially available in European countries. They observed deviations from the declared CBD content in the range of 4.44 % to 38.14 % (w/w) for 14 of the 15 analysed samples. The unlabelled sample had the lowest total CBD content with 0.24 % (w/w). Rahman et al. [41] reported similar findings for the analysis of 69 Malaysian e-liquids for their nicotine concentration. They found that more than 85 % of analysed samples were inconsistent with the labelled nicotine concentration and argued that this is due to a lack of manufacturing guidelines. Further, Palazzolo et al. [42] reported a discrepancy of labelled nicotine concentration and measured concentrations in the range of 74 to 110 % in five US e-liquids. Peace et al. [43] found in a previous study for 30 % of tested e-liquids in the United States a relative difference between detected concentrations and labelled nicotine concentrations of > plus 20 %. Furthermore, one of the analysed samples had a labelled concentration of < minus 40 % for THC and < minus 50 % for CBD, respectively [43].

According the directive 2014/40/EU of the European Parliament and of the European Council from April 2014, the emission levels of a nicotine cigarette must not be higher than 1 mg of nicotine per cigarette. It is regulated that concentrations need to be determined according to ISO 10315 [44] in the smoke condensate – not in the cigarette itself. Furthermore, it is stated, that nicotine-containing liquids are allowed to have a maximum nicotine concentration of 20 mg/mL (in the liquid). This means, that

for nicotine cigarettes and nicotine containing e-liquids different categories of maximum nicotine concentrations are considered, which makes it difficult to compare and draw conclusions from determined concentrations.

In Switzerland e-liquids are categorized according to the Swiss food legislation as commodities which are in contact with the mucosa [45]. It is regulated that commodities of this category cannot contain substances with a pharmacological effect such as nicotine or disinfectant [46]. Hence, the supplementation of e-liquids with CBD in pharmacological effective concentrations is forbidden. Since it is unclear, which dose has a pharmacological effect, and due to this wording in the Food and Commodity Law Directory it is currently not clear which concentrations of CBD can be added to e-liquids, hence CBD e-liquids are more or less unregulated in Switzerland. As a result, there are no quality specifications and requirements for manufacturers, and no regulations, how CBD-containing products have to be labelled. Therefore, it is hard to categorize the discrepancies observed for the determined and labelled CBD content. We decided to choose ± 10 % (relative) as an arbitrary "tolerable" level of deviation from the labelled CBD content for this study.

In all tested e-liquids the THC concentration was below 0.2 %, which is the maximum legal THC concentration in the European Union. It can be stipulated that this concentration was chosen in order to be able to sell the e-liquids not only in Switzerland but also the European Union.

Conclusion

The qualitative analysis could identify in total ten phytocannabinoids including the quantified analytes, six natural plant compounds and five flavours. For all 20 analysed e-liquids the matrix consisted of 1,2-propanediol and glycerol. Our results show that all investigated e-liquids met the required Swiss legal regulations of a THC content lower than 1 % and had in fact contents lower than 0.2 % (w/w), which is the maximal allowed THC concentration in the European Union. Even tough earlier publications reported on e-liquids containing synthetic cannabinoids, none of the analysed samples contained any New Psychoactive Substance (NPS). However, manufacturer information on the label and packaging varied drastically from the determined CBD content in a range of 45.9 % to 117.9 %. Furthermore, variations were observed for all five investigated producers. It is unclear how this production error occurs because for four out of five manufacturers at least one sample contained the correct labelled CBD within our defined tolerance limits of ± 10% (relative). In accordance with previous literature the two e-liquids without specification of the CBD content contained the lowest total CBD concentration (0.182 and 0.348 % (w/w)).

From our results, it can be stipulated that the e-liquids available on the Swiss market vary in quality concerning their CBD content and that consumers cannot rely on the manufacturer's information.

References

[1] Confederation, T.F.A.o.t.S., Federal Act on Narcotics and Psychotropic Substances, in: E.D.f. Inneres (Ed.) 1951.
[2] The, F.D.o.H.A., Swiss Ordinance on the Lists of Narcotics, Psychotropic Substances, Precursor and Auxiliary Chemicals. <https://www.admin.ch/opc/de/classified-compilation/20101220/index.html>, (accessed 25.09.2019.).
[3] Elsohly, M.A. Slade, D., Chemical constituents of marijuana: the complex mixture of natural cannabinoids, Life sciences 78(5) (2005) 539-48. https://doi.org/10.1016/j.lfs.2005.09.011
[4] Andre, C.M. Hausman, J.F. Guerriero, G., Cannabis sativa: The Plant of the Thousand and One Molecules, Frontiers in plant science 7 (2016) 19. https://doi.org/10.3389/fpls.2016.00019
[5] Costa, B., On the pharmacological properties of Delta9-tetrahydrocannabinol (THC), Chem Biodivers 4(8) (2007) 1664-77. https://doi.org/10.1002/cbdv.200790146
[6] Morrison, P.D. Zois, V. McKeown, D.A. Lee, T.D. Holt, D.W. Powell, J.F. Kapur, S. Murray, R.M., The acute effects of synthetic intravenous Delta9-tetrahydrocannabinol on psychosis, mood and

cognitive functioning, Psychol Med 39(10) (2009) 1607-16. https://doi.org/10.1017/S0033291709005522
[7] Freeman, T.P. Winstock, A.R., Examining the profile of high-potency cannabis and its association with severity of cannabis dependence, Psychol Med 45(15) (2015) 3181-9. https://doi.org/10.1017/S0033291715001178
[8] Ohlsson, A. Lindgren, J.E. Wahlen, A. Agurell, S. Hollister, L.E. Gillespie, H.K., Plasma delta-9 tetrahydrocannabinol concentrations and clinical effects after oral and intravenous administration and smoking, Clin Pharmacol Ther 28(3) (1980) 409-16. https://doi.org/10.1038/clpt.1980.181
[9] Pisanti, S. Malfitano, A.M. Ciaglia, E. Lamberti, A. Ranieri, R. Cuomo, G. Abate, M. Faggiana, G. Proto, M.C. Fiore, D. Laezza, C. Bifulco, M., Cannabidiol: State of the art and new challenges for therapeutic applications, Pharmacology & therapeutics 175 (2017) 133-150. https://doi.org/10.1016/j.pharmthera.2017.02.041
[10] Korfei, M., The underestimated danger of E-cigarettes - also in the absence of nicotine, Respir Res 19(1) (2018) 159. https://doi.org/10.1186/s12931-018-0870-4
[11] Giroud, C. de Cesare, M. Berthet, A. Varlet, V. Concha-Lozano, N. Favrat, B., E-Cigarettes: A Review of New Trends in Cannabis Use, Int J Environ Res Public Health 12(8) (2015) 9988-10008. https://doi.org/10.3390/ijerph120809988
[12] Poklis, J.L. Mulder, H.A. Peace, M.R., The unexpected identification of the cannabimimetic, 5F-ADB, and dextromethorphan in commercially available cannabidiol e-liquids, Forensic science international 294 (2019) e25-e27. https://doi.org/10.1016/j.forsciint.2018.10.019
[13] Angerer, V. Franz, F. Moosmann, B. Bisel, P. Auwarter, V., 5F-Cumyl-PINACA in 'e-liquids' for electronic cigarettes: comprehensive characterization of a new type of synthetic cannabinoid in a trendy product including investigations on the in vitro and in vivo phase I metabolism of 5F-Cumyl-PINACA and its non-fluorinated analog Cumyl-PINACA, Forensic Toxicol 37(1) (2019) 186-196. https://doi.org/10.1007/s11419-018-0451-8
[14] Munster-Muller, S. Matzenbach, I. Knepper, T. Zimmermann, R. Putz, M., Profiling of synthesis-related impurities of the synthetic cannabinoid Cumyl-5F-PINACA in seized samples of e-liquids via multivariate analysis of UHPLC-MS(n) data, Drug testing and analysis (2019). https://doi.org/10.1002/dta.2673
[15] Flahault, A. Etter, J.F., Electronic cigarettes: it is urgent to promote them to save lives, Int J Public Health 59(5) (2014) 681-2. https://doi.org/10.1007/s00038-014-0597-z
[16] Earleywine, M. Barnwell, S.S., Decreased respiratory symptoms in cannabis users who vaporize, Harm Reduct J 4 (2007) 11. https://doi.org/10.1186/1477-7517-4-11
[17] Lerner, C.A. Sundar, I.K. Yao, H. Gerloff, J. Ossip, D.J. McIntosh, S. Robinson, R. Rahman, I., Vapors produced by electronic cigarettes and e-juices with flavorings induce toxicity, oxidative stress, and inflammatory response in lung epithelial cells and in mouse lung, PloS one 10(2) (2015) e0116732. https://doi.org/10.1371/journal.pone.0116732
[18] Staudt, M.R. Salit, J. Kaner, R.J. Hollmann, C. Crystal, R.G., Altered lung biology of healthy never smokers following acute inhalation of E-cigarettes, Respir Res 19(1) (2018) 78. https://doi.org/10.1186/s12931-018-0778-z
[19] Knowles, H. Sun, L.H., What we know about the mysterious vaping-linked illness and deaths. <https://www.washingtonpost.com/health/2019/09/07/what-we-know-about-mysterious-vaping-linked-illnesses-deaths/>, 2019 (accessed 07.10.2019.2019).
[20] Hswen, Y. Brownstein, J.S., Real-Time Digital Surveillance of Vaping-Induced Pulmonary Disease, N Engl J Med (2019). https://doi.org/10.1056/NEJMc1912818
[21] Legislatures, N.C.o.S., Marijuana Overview. <http://www.ncsl.org/research/civil-and-criminal-justice/marijuana-overview.aspx>, 2019 (accessed 07.10.2019.2019).
[22] Eberhard, F., Zweiter Schweizer E-Zigig-Raucher lag mit Vergiftung im Spital. <https://www.blick.ch/news/schweiz/schon-47-tote-in-den-usa-zweiter-schweizer-e-zigi-raucher-lag-mit-vergiftung-im-spital-id15630428.html>, 2019 (accessed 26.11.2019).
[23] Hadener, M. Konig, S. Weinmann, W., Quantitative determination of CBD and THC and their acid precursors in confiscated cannabis samples by HPLC-DAD, Forensic science international 299 (2019) 142-150. https://doi.org/10.1016/j.forsciint.2019.03.046
[24] Gertsch, J. Leonti, M. Raduner, S. Racz, I. Chen, J.Z. Xie, X.Q. Altmann, K.H. Karsak, M. Zimmer, A., Beta-caryophyllene is a dietary cannabinoid, Proceedings of the National Academy of

Sciences of the United States of America 105(26) (2008) 9099-104. https://doi.org/10.1073/pnas.0803601105
[25] Mockute, D. Bernotiene, G. Judzentiene, A., The essential oil of Origanum vulgare L. ssp. vulgare growing wild in vilnius district (Lithuania), Phytochemistry 57(1) (2001) 65-9. https://doi.org/10.1016/s0031-9422(00)00474-x
[26] Jayaprakasha, G.K. Jagan Mohan Rao, L. Sakariah, K.K., Volatile constituents from Cinnamomum zeylanicum fruit stalks and their antioxidant activities, J Agric Food Chem 51(15) (2003) 4344-8. https://doi.org/10.1021/jf034169i
[27] Malingre, T. Hendriks, H. Batterman, S. Bos, R. Visser, J., The essential oil of Cannabis sativa, Planta Med 28(1) (1975) 56-61. https://doi.org/10.1055/s-0028-1097829
[28] Ferreira, F.M. Palmeira, C.M. Oliveira, M.M. Santos, D. Simoes, A.M. Rocha, S.M. Coimbra, M.A. Peixoto, F., Nerolidol effects on mitochondrial and cellular energetics, Toxicol In Vitro 26(2) (2012) 189-96. https://doi.org/10.1016/j.tiv.2011.11.009
[29] Chan, W.K. Tan, L.T. Chan, K.G. Lee, L.H. Goh, B.H., Nerolidol: A Sesquiterpene Alcohol with Multi-Faceted Pharmacological and Biological Activities, Molecules 21(5) (2016). https://doi.org/10.3390/molecules21050529
[30] Inoue, Y. Shiraishi, A. Hada, T. Hirose, K. Hamashima, H. Shimada, J., The antibacterial effects of terpene alcohols on Staphylococcus aureus and their mode of action, FEMS Microbiol Lett 237(2) (2004) 325-31. https://doi.org/10.1016/j.femsle.2004.06.049
[31] Navarro-Moll, M.C. Romero, M.C. Montilla, M.P. Valero, A., In vitro and in vivo activity of three sesquiterpenes against L(3) larvae of Anisakis type I, Exp Parasitol 127(2) (2011) 405-8. https://doi.org/10.1016/j.exppara.2010.09.008
[32] Lee, K. Lee, J.H. Kim, S.I. Cho, M.H. Lee, J., Anti-biofilm, anti-hemolysis, and anti-virulence activities of black pepper, cananga, myrrh oils, and nerolidol against Staphylococcus aureus, Appl Microbiol Biotechnol 98(22) (2014) 9447-57. https://doi.org/10.1007/s00253-014-5903-4
[33] Vinholes, J. Goncalves, P. Martel, F. Coimbra, M.A. Rocha, S.M., Assessment of the antioxidant and antiproliferative effects of sesquiterpenic compounds in in vitro Caco-2 cell models, Food Chem 156 (2014) 204-11. https://doi.org/10.1016/j.foodchem.2014.01.106
[34] Lima, D.K. Ballico, L.J. Rocha Lapa, F. Goncalves, H.P. de Souza, L.M. Iacomini, M. Werner, M.F. Baggio, C.H. Pereira, I.T. da Silva, L.M. Facundo, V.A. Santos, A.R., Evaluation of the antinociceptive, anti-inflammatory and gastric antiulcer activities of the essential oil from Piper aleyreanum C.DC in rodents, J Ethnopharmacol 142(1) (2012) 274-82. https://doi.org/10.1016/j.jep.2012.05.016
[35] Klopell, F.C. Lemos, M. Sousa, J.P. Comunello, E. Maistro, E.L. Bastos, J.K. de Andrade, S.F., Nerolidol, an antiulcer constituent from the essential oil of Baccharis dracunculifolia DC (Asteraceae), Z Naturforsch C J Biosci 62(7-8) (2007) 537-42. https://doi.org/10.1515/znc-2007-7-812
[36] Cornwell, P.A. Barry, B.W., Sesquiterpene components of volatile oils as skin penetration enhancers for the hydrophilic permeant 5-fluorouracil, J Pharm Pharmacol 46(4) (1994) 261-9. https://doi.org/10.1111/j.2042-7158.1994.tb03791.x
[37] Bundesrat, D., Gesetz über den Verkehr mit Betäubungsmitteln in: B.f.J.u. Verbraucherschutz (Ed.) BGBl. I S. 1202, Bundesamt für Justiz, 2019, p. 56.
[38] Peace, M.R. Butler, K.E. Wolf, C.E. Poklis, J.L. Poklis, A., Evaluation of Two Commercially Available Cannabidiol Formulations for Use in Electronic Cigarettes, Frontiers in pharmacology 7 (2016) 279. https://doi.org/10.3389/fphar.2016.00279
[39] Bonn-Miller, M.O. Loflin, M.J.E. Thomas, B.F. Marcu, J.P. Hyke, T. Vandrey, R., Labeling Accuracy of Cannabidiol Extracts Sold Online, JAMA 318(17) (2017) 1708-1709. https://doi.org/10.1001/jama.2017.11909
[40] Pavlovic, R. Nenna, G. Calvi, L. Panseri, S. Borgonovo, G. Giupponi, L. Cannazza, G. Giorgi, A., Quality Traits of "Cannabidiol Oils": Cannabinoids Content, Terpene Fingerprint and Oxidation Stability of European Commercially Available Preparations, Molecules 23(5) (2018). https://doi.org/10.3390/molecules23051230
[41] Rahman, A. Mohamed, M.H.N. Mahmood, S., Nicotine Estimations in Electronic Cigarette E-Liquids Among Malaysian Marketed Samples, Analytical Chemistry Letters 8(1) (2018) 54-62. https://doi.org/10.1080/22297928.2017.1400920

[42] Palazzolo, D. Nelson, J.M. Hudson, Z., The Use of HPLC-PDA in Determining Nicotine and Nicotine-Related Alkaloids from E-Liquids: A Comparison of Five E-Liquid Brands Purchased Locally, Int J Environ Res Public Health 16(17) (2019). https://doi.org/10.3390/ijerph16173015
[43] Peace, M.R. Stone, J.W. Poklis, J.L. Turner, J.B. Poklis, A., Analysis of a Commercial Marijuana e-Cigarette Formulation, Journal of analytical toxicology 40(5) (2016) 374-8. https://doi.org/10.1093/jat/bkw021
[44] union, E.P.a.t.C.o.t.E., DIRECTIVE 2014/40/EU OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 3 April 2014 on the approximation of the laws, regulations and administrative provisions of the Member States concerning the manufacture, presentation and sale of tobacco and related products and repealing Directive 2001/37/EC, L127/1, Official Journal of the European Union, 2014, p. 38.
[45] Constitution, F., Federal Act on Foodstuffs and Utility Articles, in: T.F.A.o.t.S. Confederation (Ed.) 817.0, 2017.
[46] Council, S.F., Food and commodity law directory, in: S.F. Council (Ed.) 817.02, 2019, p. 44.